PATENT
600-1-158N DIV

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

APPLICANT :	ROBERT H. MASURE *ET AL.*		
SERIAL NO. :	UNASSIGNED	EXAMINER :	UNKNOWN
FILED :	HEREWITH	ART UNIT :	UNKNOWN
FOR :	CHOLINE BINDING PROTEINS FOR ANTI-PNEUMOCOCCAL VACCINES		

VIA EXPRESS MAIL NO. EL684490894US
DATE OF DEPOSIT: APRIL 9, 2001

PRELIMINARY AMENDMENT

BOX PATENT APPLICATION
ASSISTANT COMMISSIONER FOR PATENTS
WASHINGTON, D.C. 20231

Sir:

In accordance with Rule 111 of the Rules of Practice please consider the following amendments and remarks.

IN THE SPECIFICATION:

On page 1, prior to line 3, please insert the following new section:

- - CROSS REFERENCE TO RELATED APPLICATION

This application is a divisional of Serial No. 08/847,065, filed May 1, 1997, the subject matter of which is hereby incorporated by reference in its entirety. - -

Please replace the Sequence Listing in the original Specification with the enclosed Substitute Sequence Listing.

IN THE CLAIMS:

Please cancel Claims 1-12, 19-27, and 29-32 without prejudice.

Please add the following claims:

- - 41. An isolated nucleic acid encoding a streptococcal choline binding protein;

wherein the protein is expressed by *Streptococcus pneumoniae* and has the following characteristics:

a) choline-binding activity;

b) elution from a chromatographic column in the presence of about 10% choline;

c) being reactive with sera from patients infected or recovering from infection with the bacteria;

d) being labeled by fluorescein isothiocyanate (FITC) without requiring streptococcal lysis; and

e) comprising an amino acid sequence selected from the group consisting of SEQ ID NO:1, and SEQ ID NO: 6.

42. The isolated nucleic acid of Claim 41 that is a recombinant DNA molecule.

43. The recombinant DNA molecule of Claim 42 that is operatively linked to an expression control sequence.

44. A unicellular host transformed with the recombinant DNA molecule of Claim 43.

45. A DNA vaccine comprising the recombinant DNA molecule of Claim 43.

46. An oligonucleotide capable of screening for a nucleic acid encoding a streptococcal choline binding protein prepared from the nucleic acid of Claim 41.

47. The isolated nucleic acid of Claim 41 wherein said streptococcal choline binding protein comprises the amino acid sequence of SEQ ID NO:1.

48. An isolated nucleic acid encoding a streptococcal choline binding protein comprising the amino acid sequence of SEQ ID NO:25 or SEQ ID NO:25 comprising a conservative amino acid substitution; wherein the isolated streptococcal choline binding protein has the following characteristics:

a) choline-binding activity;

b) elution from a chromatographic column in the presence of about 10% choline; and

c) being reactive with sera from patients infected or recovering from infection with the bacteria.

49. The isolated nucleic acid of Claim 48 which comprises the nucleotide sequence of SEQ ID NO:24.

50. The isolated nucleic acid of Claim 48 that is a recombinant DNA molecule.

51. The recombinant DNA molecule of Claim 49 that is operatively linked to an expression control sequence.

52. A unicellular host transformed with the recombinant DNA molecule of Claim 51.

53. A DNA vaccine comprising the recombinant DNA molecule of Claim 51.

54. The isolated nucleic acid of Claim 48 wherein the streptococcal choline binding protein further comprises the amino acid sequence of SEQ ID NO:1.

55. An isolated nucleic acid encoding an antigenic fragment of the N-terminal region of a streptococcal choline binding protein; wherein the streptococcal choline binding protein comprises the amino acid sequence of SEQ ID NO:25 or SEQ ID NO:25 comprising a conservative amino acid substitution; and wherein the streptococcal choline binding protein has the following characteristics:

a) choline-binding activity;

b) elution from a chromatographic column in the presence of about 10% choline; and

c) being reactive with sera from patients infected or recovering from infection with the bacteria.

56. (Amended) The isolated nucleic acid of Claim 55, wherein said streptococcal choline binding protein further comprises the amino acid sequence of SEQ ID NO:1.

57. (Amended) The isolated nucleic acid of Claim 55 wherein said streptococcal choline binding protein comprises one or two lectin binding domains of the N-terminal domain of the streptococcal choline binding protein.

58. The isolated nucleic acid of Claim 55 that is a recombinant DNA molecule.

59. The recombinant DNA molecule of Claim 58 that is operatively linked to an expression control sequence.

60. A unicellular host transformed with the recombinant DNA molecule of Claim 59.

61. A DNA vaccine comprising the recombinant DNA molecule of Claim 59.

62. A nucleic acid that hybridizes to the nucleotide sequence of SEQ ID:20 and/or SEQ ID NO:24 under highly stringent hybridization conditions.

63. A nucleotide sequence that encodes a fragment of a choline binding protein consisting of the amino acid sequence of SEQ ID NO:4.

64. An isolated nucleotide sequence that encodes a streptococcal choline binding protein comprising the amino acid sequence of SEQ ID NO:19; wherein said streptococcal choline binding protein comprises enolase activity.

65. The isolated nucleotide sequence of Claim 64 which has a nucleotide sequence as depicted in SEQ ID NO:18 from nucleotide 1 through the stop codon TAA.

66. A nucleic acid that hybridizes to the nucleotide sequence of SEQ ID:14 and/or SEQ ID NO:18 under highly stringent hybridization conditions.

67. The isolated nucleic acid of Claim 66 that is a recombinant DNA molecule.

68. The recombinant DNA molecule of Claim 67 that is operatively linked to an expression control sequence.

69. A unicellular host transformed with the recombinant DNA molecule of Claim 68.

70. A DNA vaccine comprising the recombinant DNA molecule of Claim 68.

71. A method for detecting the presence of a bacterium comprising a nucleic acid encoding a streptococcal choline binding protein comprising:

(a) contacting a sample in which the presence or activity of the bacterium is suspected with the oligonucleotide of Claim 46; and

(b) detecting whether hybridization has occurred between the oligonucleotide and the nucleic acid; wherein detection of hybridization indicates that presence or activity of the bacterium in the sample.

72. A method for preventing infection with a bacterium that expresses a streptococcal choline binding protein comprising administering an immunogenically effective dose of the DNA vaccine of Claim 45 to a subject. - -

REMARKS

Applicants respectfully request entry of the foregoing amendment into the file history of the above-identified Application being filed herewith. Support for the new claims can be found throughout the Specification including in the original claims. No new matter has been entered. Claims 13-18, 28, 33-40 and newly added Claims 41-72 are pending. Early and favorable action on the pending set of Claims is earnestly solicited.

Respectfully submitted,

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